A. Exon 1

B. Exon 2

C. Exons 3 and 4

FIGURE 1

FIGURE 2

FIGURE 3

Aatll (3686) Sall (3696)
Hpal (3676) BspHI (87)

Ncol (2830)
BspHI (2719) pFerX2
Eagl (2639) (3701 bp)
SacII (2575) Exon 1 (2567)
BamHI (2515)

BspHI (1588)

Deletion of the region from
BspHI (2719) to Ncol (2830)

Aatll (3575) Sall (3585)
Hpal (3565)

BspHI/Ncol (2721)
pFerX3
(3590 bp)
Eagl (2639)
SacII (2575)
BamHI (2515)

Deletion of the IRE by
replacement of the SacII/Eagl
fragment with a linker.

Aatll (3547)
Hpal (3537) Sall (3557)

Eagl (2611)
Kpnl (2579) pFerX4
SacII (2575) (3562 bp)
BamHI (2515)
EcoRV (2508)

FIGURE 4

A.

Fer1
ATC TGT CCA TG GTGAGTGCGGCCTGGCCTTTGGCGGGGCGGAAAGAGGGTGCGGCCTGGCCTCCCTTGGGCCACTTGGTGAGCTGGCGGAGG

GTGGGTTGGGGCGTGGCCTGCTGCGGGCTTCCCCGCCTTCCAGCGCCCTTCTGGAAAATGGAGTTTGTCCGGGGTTCTTTCCAAAGGCAGGCAGCC
CTGCCGTGGCAAGTCTGAGCACCTAGCGCTTTGTGGCTCCTGCATAGACCAGGCACGTCATAACACCCGTGTTTTGAAGCCTTAGGGCTGTACAAC
TGTCAGCCTCTCCAATCAACCCTGCAGTTAGGTGCATTTTCCTGCACTCTCGTCCCCTCCGGTCACATGGCCTGCAGGCTTCTCTGTTTGGGTGTA
CATCCAGCTCCAGTTCCTCTGACTATGGCGGGTCTGCTTGGTCATGGTGTGGAATGGCAGCCCTGGGGCTTGGTACAAAGAGGCTTATCTCTTGTG
AACTTACTCTAACCACTTCTGAAGCAGCGGCCTCTACATCTCTGCTTATCACAGAGCCTCACTTGCATTGAAACTTATCGCTAGGAATCTCCCCTT
CTGTAATCACCCTGACCTTGCCAAGGCATCTAGAGTACTGTACGTTTTTAATTTTTATTTTGCACCAGTTGTTGCTTACTAACAGAAGTAGTAGGT
AACATACTTGTTGGAAAAAGCCCACGGTTGGGAAAAAAACCATTATCGTGGAATACAAATACACTGAGTGCCTAAAACTGAAAATCAAAGCTTCTCC

                                         Hpal
CAATGTATTTGTGCTAAAATACAATGCCCTCAGTTCTTAACCAGGTAATCAGCAGTTGGCTGTCTAGCTGAAAACCTTGAGACCTTGTGTTAACCA
TTTTTTTTATTTAACATGATTGTTGAAGGAGAGAATTGACCTCCCAATGTAGGGCACTTTAGCACCCCCCCTCTCAGACAAATAGATATGGCCTTG
GCTTAAAGTTTTTTCTCTGCACTAATGTGGAGCCATAGAACCCTTGATAAAGCCAAGTCCCAAGTTTGTTTTCCCATCCTTACTTTAAAGGCCAAG

                                        Exon 2
TAGGGTGACAAACAGCCTTTACCACCATTGCATCTGCCTTGCTGTGGGGATCAATAACAAATACCCTTTCCACTTTCAG TCT TGT TAT TTT
                                                    Ser Cys Tyr Phe

GAC CGG GAT GAT GTG GCC CTG AAG AAC TTT GCC AAA TAC TTT CTC CAT CAA TCT CAT GAA GAG AGG GAA CAT
▶Asp Arg Asp Asp Val Ala Leu Lys Asn Phe Ala Lys Tyr Phe Leu His Gln Ser His Glu Glu Arg Glu His
GCT GAG AAA CTG ATG AAG CTG CAG AAC CAG CGA GGT GGA CGA ATC TTC CTG CAG GAT ATC AAG GTAAGTAGACTA
▶Ala Glu Lys Leu Met Lys Leu Gln Asn Gln Arg Gly Gly Arg Ile Phe Leu Gln Asp Ile Lys
TGGGACTGCGTTAAATGAGCAGTNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNN
NNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNCTGCAGATGAATTGACATG
           Exon 3
TTTCTTTGATTCAG AAA CCT GAC CGT GAT GAC TGG GAG AGC GGG CTG AAT GCA ATG AGG TGT GCA CTG CAC TTG
                ▶Lys Pro Asp Arg Asp Asp Trp Glu Ser Gly Leu Asn Ala Met Arg Cys Ala Leu His Leu
GAA AAG AGT GTG AAT CAG TCA CTA CTG GAA CTT CAC AAA CTG GCT ACT GAC AAG AAT GAT CCC CAC GTGAGTAT
▶Glu Lys Ser Val Asn Gln Ser Leu Leu Glu Leu His Lys Leu Ala Thr Asp Lys Asn Asp Pro His
                                            Exon 4
CAGAAACACGGGGTGAGTGGAGATGATTTGCCACAGGGCTTGGGAGAGCTGACCAGTAACCCTGTCCCATGTTCTCTTTCCTAG TTA TGT GAC
                                                   ▶Leu Cys Asp
TTC ATT GAG ACG CAT TAC CTG AAT GAG CAG GTG AAA TCC ATT AAA GAA CTG GGT GAC CAC GTG ACC AAC TTA
▶Phe Ile Glu Thr His Tyr Leu Asn Glu Gln Val Lys Ser Ile Lys Glu Leu Gly Asp His Val Thr Asn Leu
CGC AAG ATG GGA GCC CCT GAA TCT GGC ATG GCA GAA TAT CTC TTT GAC AAG CAC ACC CTG GGA CAC GGT GAT
▶Arg Lys Met Gly Ala Pro Glu Ser Gly Met Ala Glu Tyr Leu Phe Asp Lys His Thr Leu Gly His Gly Asp
           Aatll (5419)
GAG AGC TAA GCTGACGTCCCCAAGGCCATGTGACTTTACTGGTCACTGAGGCAGTGCATGCATGTCAGGCTGCCTTTATCTTTTCTATAAGTT
▶Glu Ser •••

GCACCAAAACATCTGCTTAAAAGTTCTTTAATTTGTACCATTTCTTCAAATAAAGAATTTTGGTACCCAGCTCTTGTTGTGATTG
                                                      Fer4

B.

Fer1
ATC TGT CCA TG GTGAGTGCGGCCTGGCCTTTGGCGGGGCGGAAAGAGGGTGCGGCCTGGCCTCCCTTGGGCCACTTGGTGAGCTGGCGGAGGG

TGGGTTGGGGCGTGGCCTGCTGCGGGCTTCCCCGCCTTCCAGCGCCCTTCTGGAAAATGGAGTTTGTCCGGGGTTCTTTCCAAAGGCAGGCAGCCCT
GCCGTGGCAAGTCTGAGCACCTAGCGCTTTGTGGCTCCTGCATAGACCAGGCACGTCATAACACCCGTGTTTTGAAGCCTTAGGGCTGTACAACTGT
CAGCCTCTCCAATCAACCCTGCAGTTAGGTGCATTTTCCTGCACTCTCGTCCCCTCCGGTCACATGGCCTGCAGGCTTCTCTGTTTGGGTGTACATC
CAGCTCCAGTTCCTCTGACTATGGCGGGTCTGCTTGGTCATGGTGTGGAATGGCAGCCCTGGGGCTTGGTACAAAGAGGCTTATCTCTTGTGAACTT
ACTCTAACCACTTCTGAAGCAGCGGCCTCTACATCTCTGCTTATCACAGAGCCTCACTTGCATTGAAACTTATCGCTAGGAATCTCCCCTTCTGTAA
TCACCCTGACCTTGCCAAGGCATCTAGAGTACTGTACGTTTTTAATTTTTATTTTGCACCAGTTGTTGCTTACTAACAGAAGTAGTAGGTAACATAC
TTGTTGGAAAAAGCCCACGGTTGGGAAAAAAACCATTATCGTGGAATACAAATACACTGAGTGCCTAAAACTGAAAATCAAAGCTTCTCCCAATGTAT
                                        Hpal
TTGTGCTAAAATACAATGCCCTCAGTTCTTAACCAGGTAATCAGCAGTTGGCTGTCTAGCTGAAAACCTTGAGACCTTGTGTTAACCATTTTTTTTA
TTTAACATGATTGTTGAAGGAGAGAATTGACCTCCCAATGTAGGGCACTTTAGCACCCCCCCTCTCAGACAAATAGATATGGCCTTGGCTTAAAGTT
TTTTCTCTGCACTAATGTGGAGCCATAGAACCCTTGATAAAGCCAAGTCCCAAGTTTGTTTTCCCATCCTTACTTTAAAGGCCAAGTAGGGTGACAA
                                Swa-2   Notl   FN2
ACAGCCTTTACCACCATTGCATCTGCCTTGCTGTGGGGATCAATAACAAATACCCTTTCCACTTTCAGCTGCTAGCGGCCGCGCTGACGT
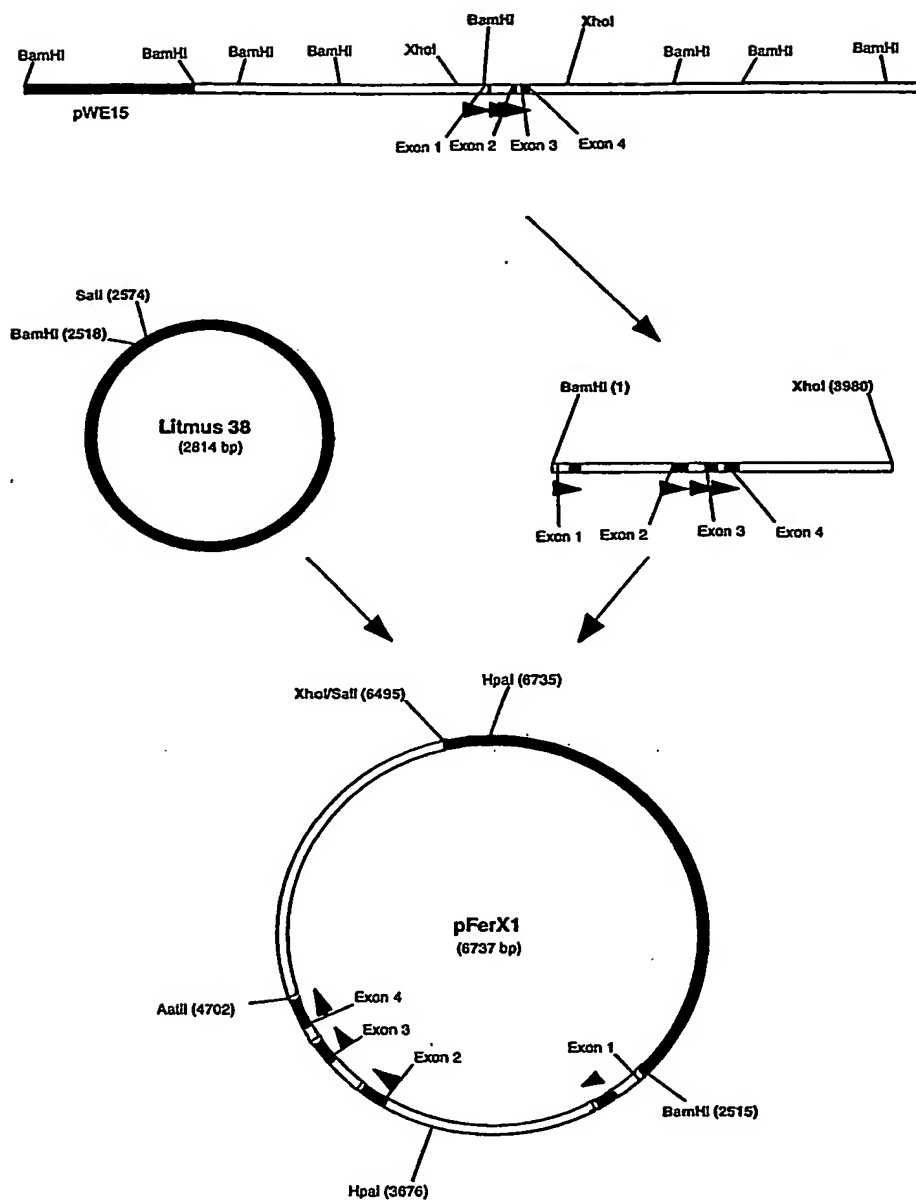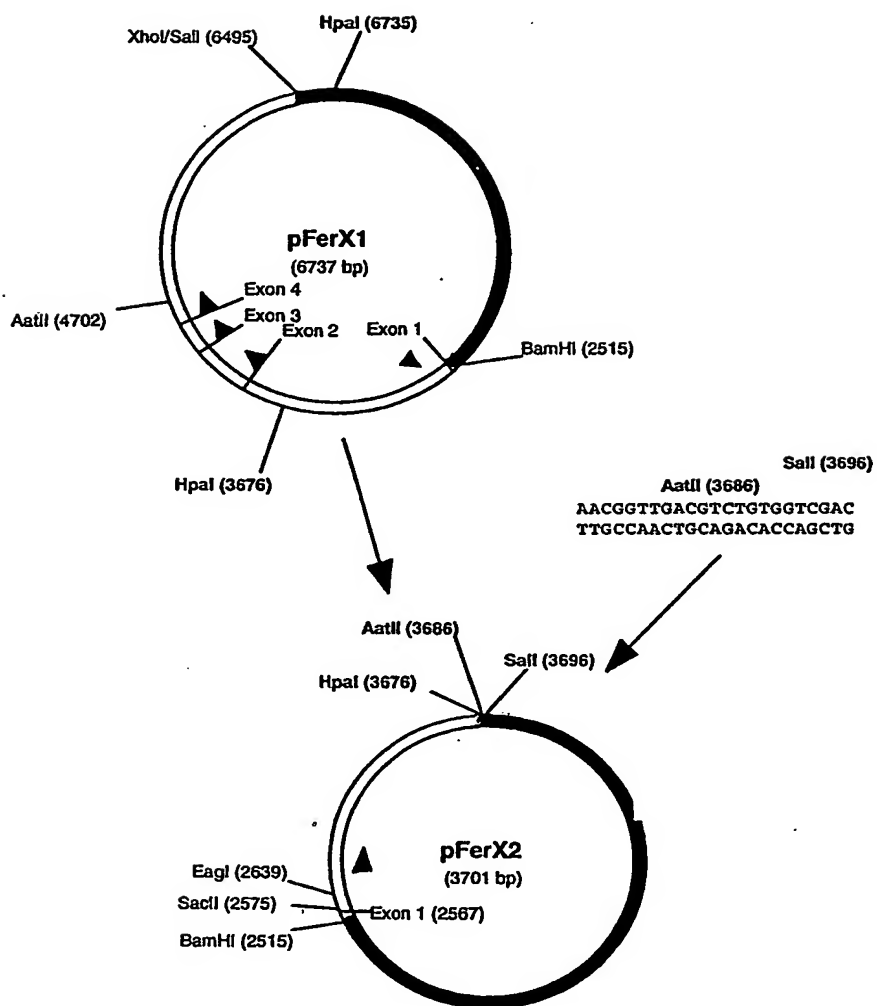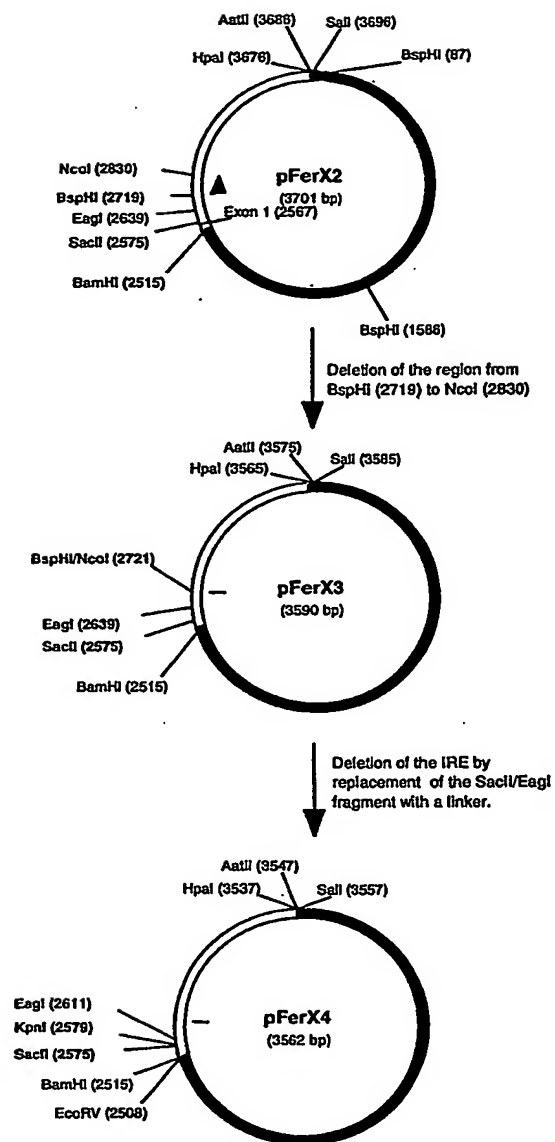
FIGURE 5A

C.

FN1   Swa-1 NotI   AatII

ACTTTCAGCTGCTAGCGGCCGCGCTGACGTCCCCAAGGCCATGTGACTTTACTGGTCACTGAGGCAGTGCATGCATGTCAGGCTGCCTTTATCTTT

Fer4

TCTATAAGTTGCACCAAAACATCTGCTTAAAAGTTCTTTAATTTGTACCATTTCTTCAAATAAAGAATTTTGGTACCCAGCTCTTGTTGTGATTG

D.

Fer1

ATC TGT CCA TG GTGAGTGCGGCCTGGCCTTTGGCGGGGCGGAAAGAGGGTGCGGCCTGGCCTCCCTTGGGCCACTTGGTGAGCTGGCGGAGGG

TGGGTTGGGGCGTGGCCTGCTGCGGGCTTCCCCGCCTTCCAGCGCCCTTCTGGAAAATGGAGTTTGTCCGGGGTTCTTTCCAAAGGCAGGCAGCCCT
GCCGTGGCAAGTCTGAGCACCTAGCGCTTTGTGGCTCCTGCATAGACCAGGCACGTCATAACACCCGTGTTTTGAAGCCTTAGGGCTGTACAACTGT
CAGCCTCTCCAATCAACCCTGCAGTTAGGTGCATTTTCCTGCACTCTCGTCCCCTCCGGTCACATGGCCTGCAGGCTTCTCTGTTTGGGTGTACATC
CAGCTCCAGTTCCTCTGACTATGGCGGGTCTGCTTGGTCATGGTGTGGAATGGCAGCCCTGGGGCTTGGTACAAAGAGGCTTATCTCTTGTGAACTT
ACTCTAACCACTTCTGAAGCAGCGGCCTCTACATCTCTGCTTATCACAGAGCCTCACTTGCATTGAAACTTATCGCTAGGAATCTCCCCTTCTGTAA
TCACCCTGACCTTGCCAAGGCATCTAGAGTACTGTACGTTTTTAATTTTTATTTTGCACCAGTTGTTGCTTACTAACAGAAGTAGTAGGTAACATAC
TTGTTGGAAAAAGCCCACGGTTGGGAAAAAAACCATTATCGTGGAATACAAATACACTGAGTGCCTAAAACTGAAAATCAAAGCTTCTCCCAATGTAT

Hpal
TTGTGCTAAAATACAATGCCCTCAGTTCTTAACCAGGTAATCAGCAGTTGGCTGTCTAGCTGAAAACCTTGAGACCTTGTGTTAACCATTTTTTTTA
TTTAACATGATTGTTGAAGGAGAGAATTGACCTCCCAATGTAGGGCACTTTAGCACCCCCCCTCTCAGACAAATAGATATGGCCTTGGCTTAAAGTT
TTTTCTCTGCACTAATGTGGAGCCATAGAACCCTTGATAAAGCCAAGTCCCAAGTTTGTTTCCCATCCTTACTTTAAAGGCCAAGTAGGGTGACAA

Swa-1 Swal   Swa-2 NotI   AatII
ACAGCCTTTACCACCATTGCATCTGCCTTGCTGTGGGGATCAATAACAAATACCCTTTCCATTTAAATCTGCTAGCGGCCGCTGACGTCCCCAAGGC

CATGTGACTTTACTGGTCACTGAGGCAGTGCATGCATGTCAGGCTGCCTTTATCTTTTCTATAAGTTGCACCAAAACATCTGCTTAAAAGTTCTTTA

Fer4
ATTTGTACCATTTCTTCAAATAAAGAATTTTGGTACCCAGCTCTTGTTGTGATTG
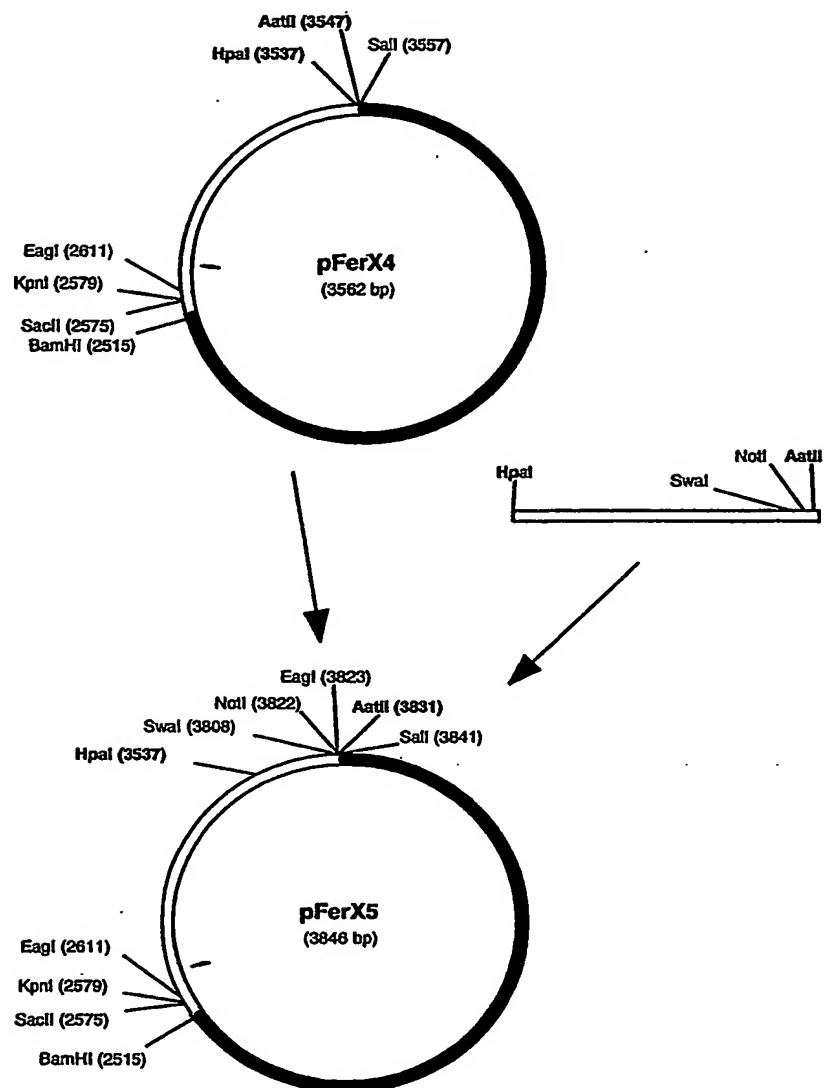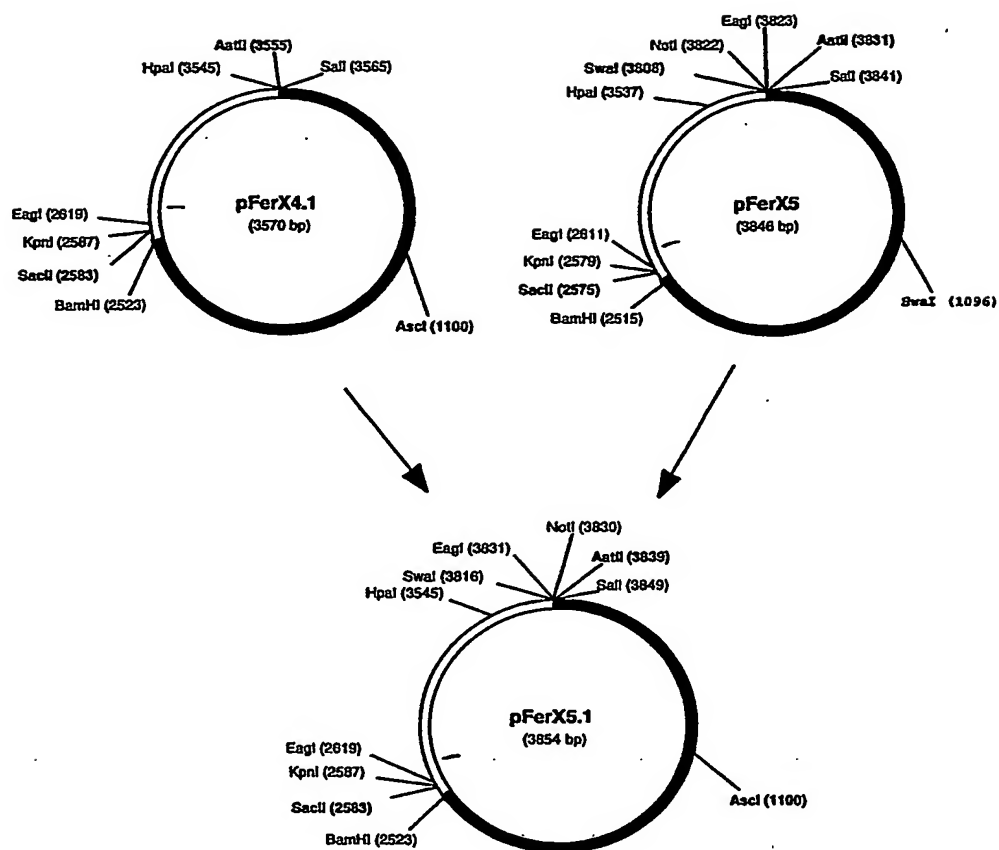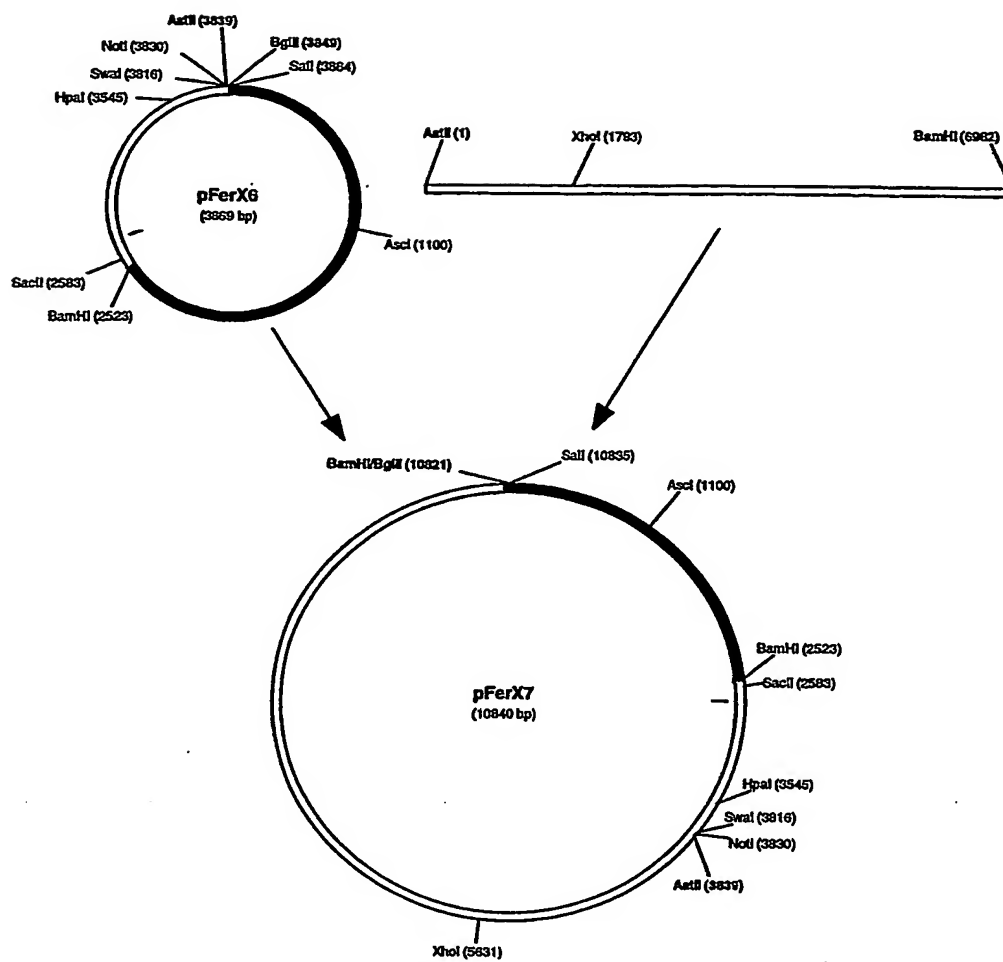
FIGURE 5 β

FIGURE 6

FIGURE 7

FIGURE 8

FIGURE 9
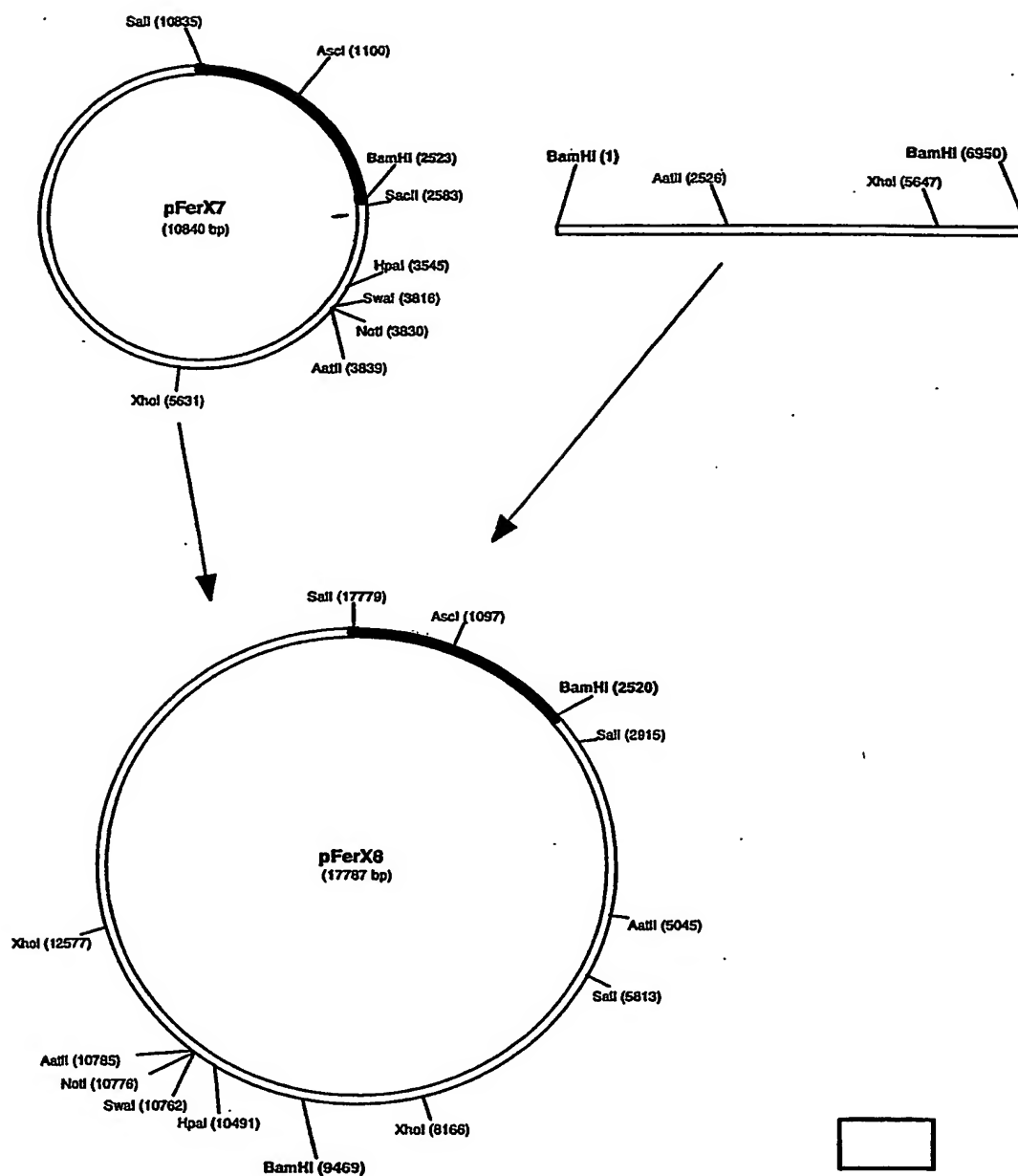
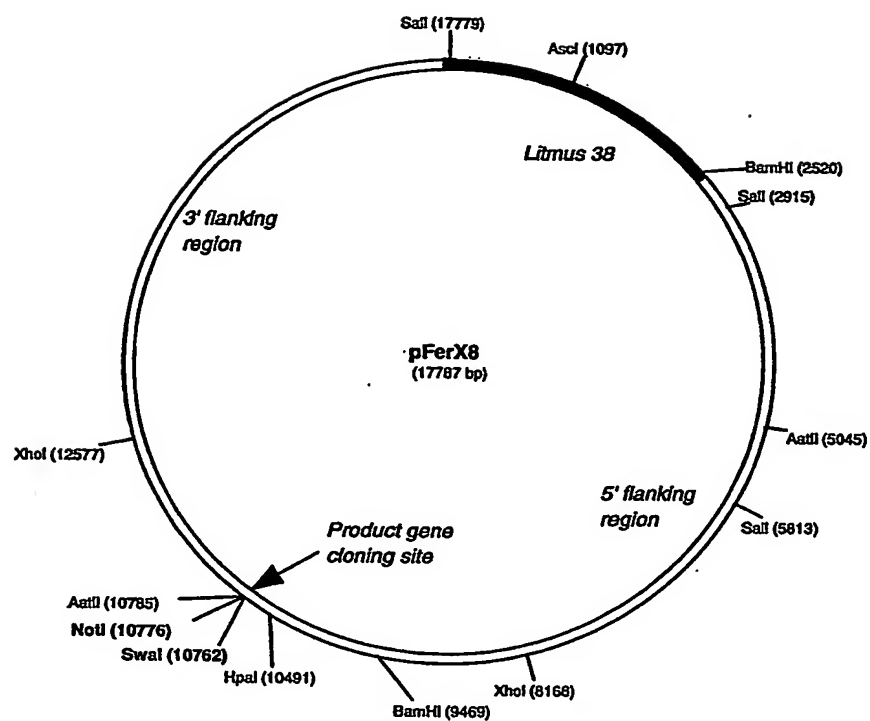**FIGURE 10**

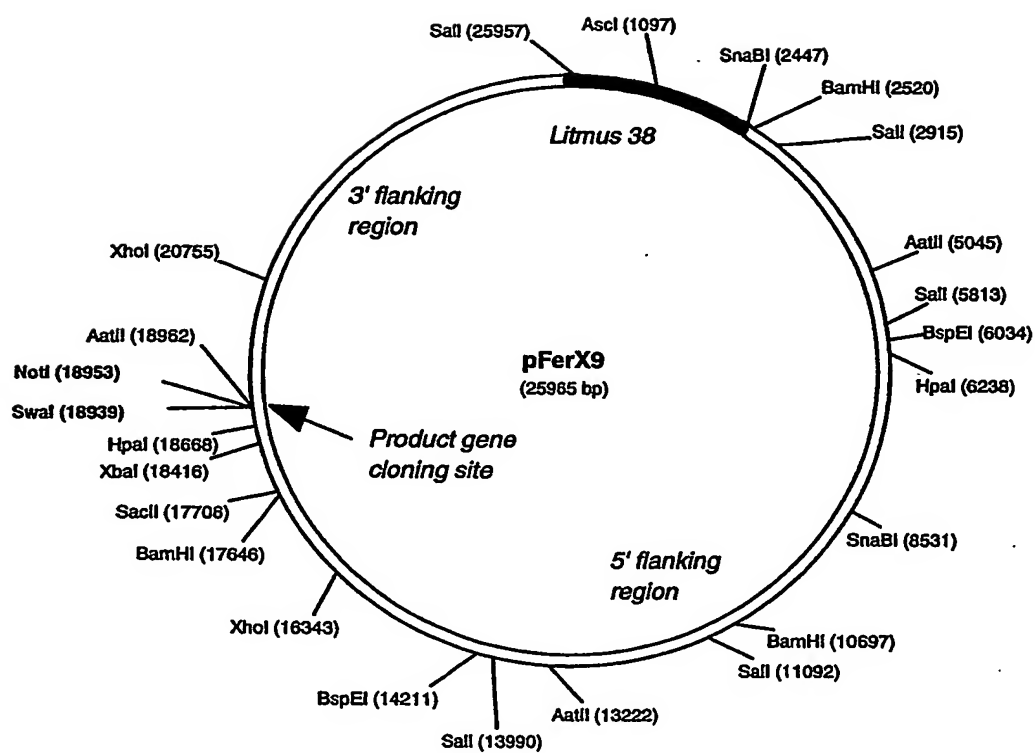FIGURE 11

SacII

BamHI (9469)   CAP (9521)   KpnI (9533)

GGATCCCGCTATAAAGTGCGGCCCGCTGGTCCCTACGCCAGACGTTCTCGCCCAGAGTCGCCGCGGTACCGGTGCTCG

ACCCCTCCGACCCCCGTCCGGCCGCTTTGAGCCTGAGCCCTTTGCAACTTCGTCGCTCCGCCGCTCCAGCGTCGCCTC

CGCGCCTCGCCCAGCCGCCATC ATG gtgagtgcggcctggcctttggcggggcggaaagagggtgcggcctggcct
                        ►Met

ccccttgggccacttggtgagctggcggagggtgggttggggcgtggcctgctgcgggcttccccgccttccagcgccc

ttctggaaaatggagtttgtccggggttctttccaaaggcaggcagccctgccgtggcaagtctgagcacctagcgct

ttgtggctcctgcatagaccaggcacgtcataacacccgtgttttgaagccttagggctgtacaactgtcagcctctc

caatcaaccctgcagttaggtgcattttcctgcactctcgtccctccggtcacatggcctgcaggcttctctgtttg

ggtgtacatccagctccagttcctctgactatggcgggtctgcttggtcatggtgtggaatggcagccctggggcttg

gtacaaagaggcttatctcttgtgaacttactctaaccacttctgaagcagcggcctctacatctctgcttatcacag

agcctcacttgcattgaaacttatcgctaggaatctcccccttctgtaatcaccctgaccttgccaaggcatctagagt

actgtacgttttaattttttatttgcaccagttgttgcttactaacagaagtagtaggtaacatacttgttggaaaa

agcccacggttgggaaaaaaccattatcgtggaatacaaatacactgagtgcctaaaactgaaaatcaaagcttctcc

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agcaccccccctctcagacaaatagatatggccttggcttaaagttttttctctgcactaatgtggagccatagaacc

cttgataaagccaagtcccaagtttgtttcccatccttactttaaaggccaagtagggtgacaaacagcctttacca

AatII (10785)

Swa I (10762)   NotI (10776)

ccattgcatctgccttgctgtggggatcaataacaaatacccttccatttAAATCTGCTAGCGGCCGCTGACGTCCC

CAAGGCCATGTGACTTTACTGGTCACTGAGGCAGTGCATGCATGTCAGGCTGCCTTTATCTTTTCTATAAGTTGCACC

KpnI (10927)

AAAACATCTGCTTAAAAGTTCTTTAATTTGTACCATTTCTTCAAATAAAGAATTTTGGTACCCAGCTCTTGTTGTGAT

TGAGGATGAGCGCACCAGCTTCCCTTGCGTCGGCTATACTAACCACACTGCA

FIGURE 12

FIGURE 13
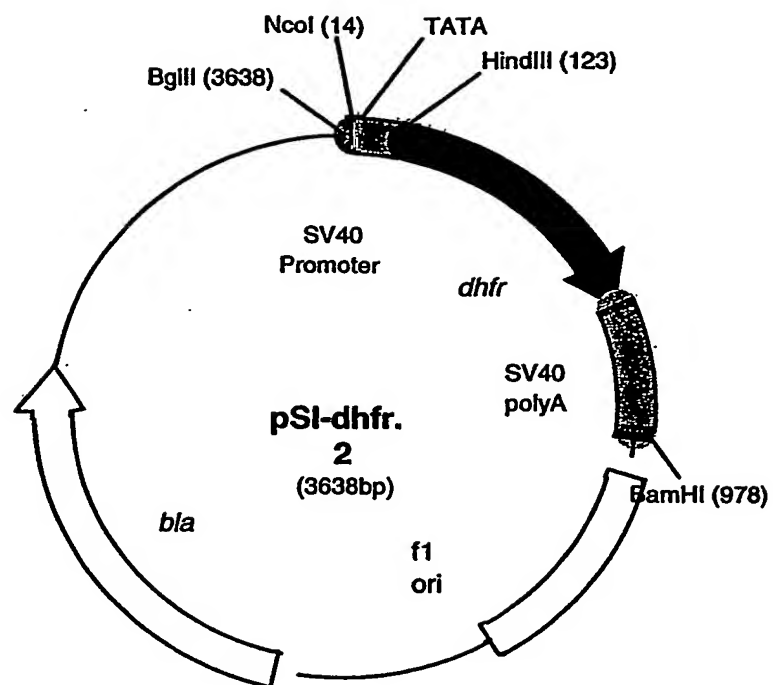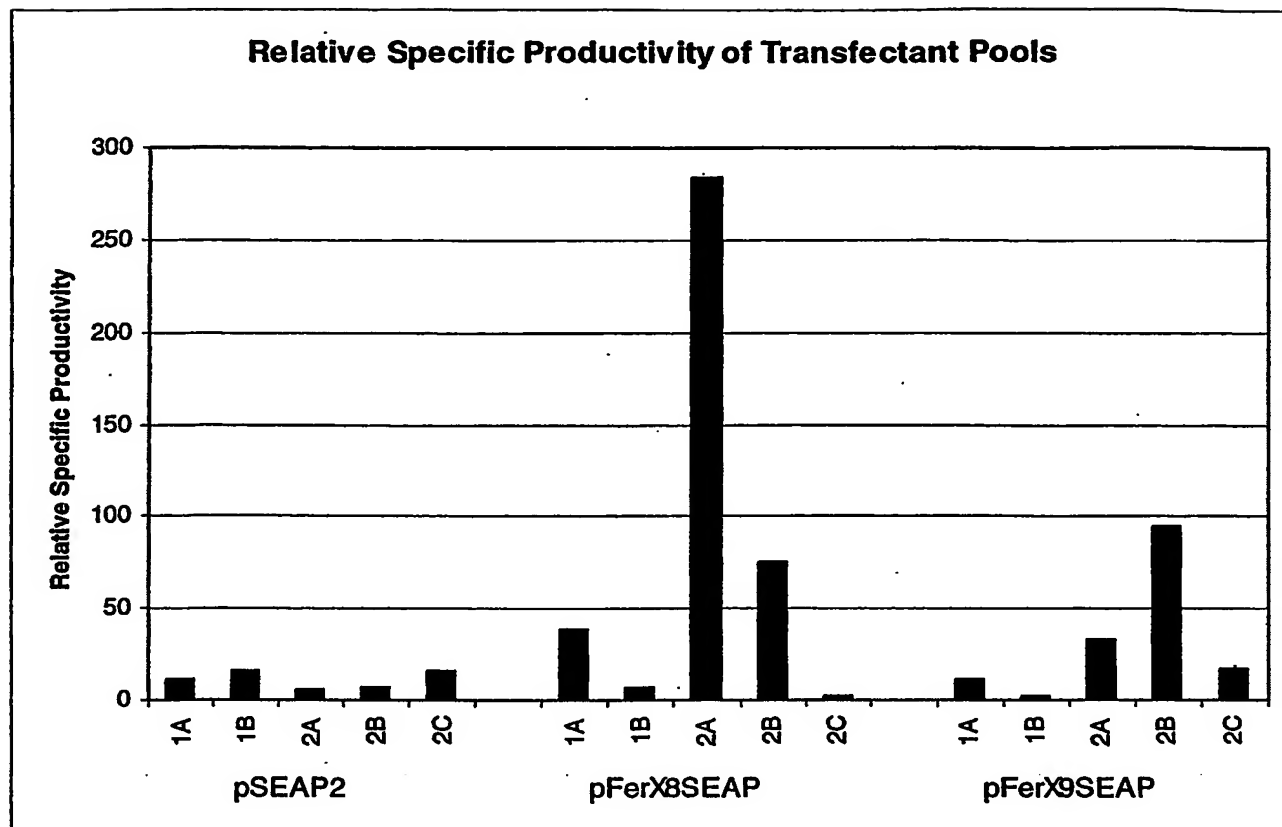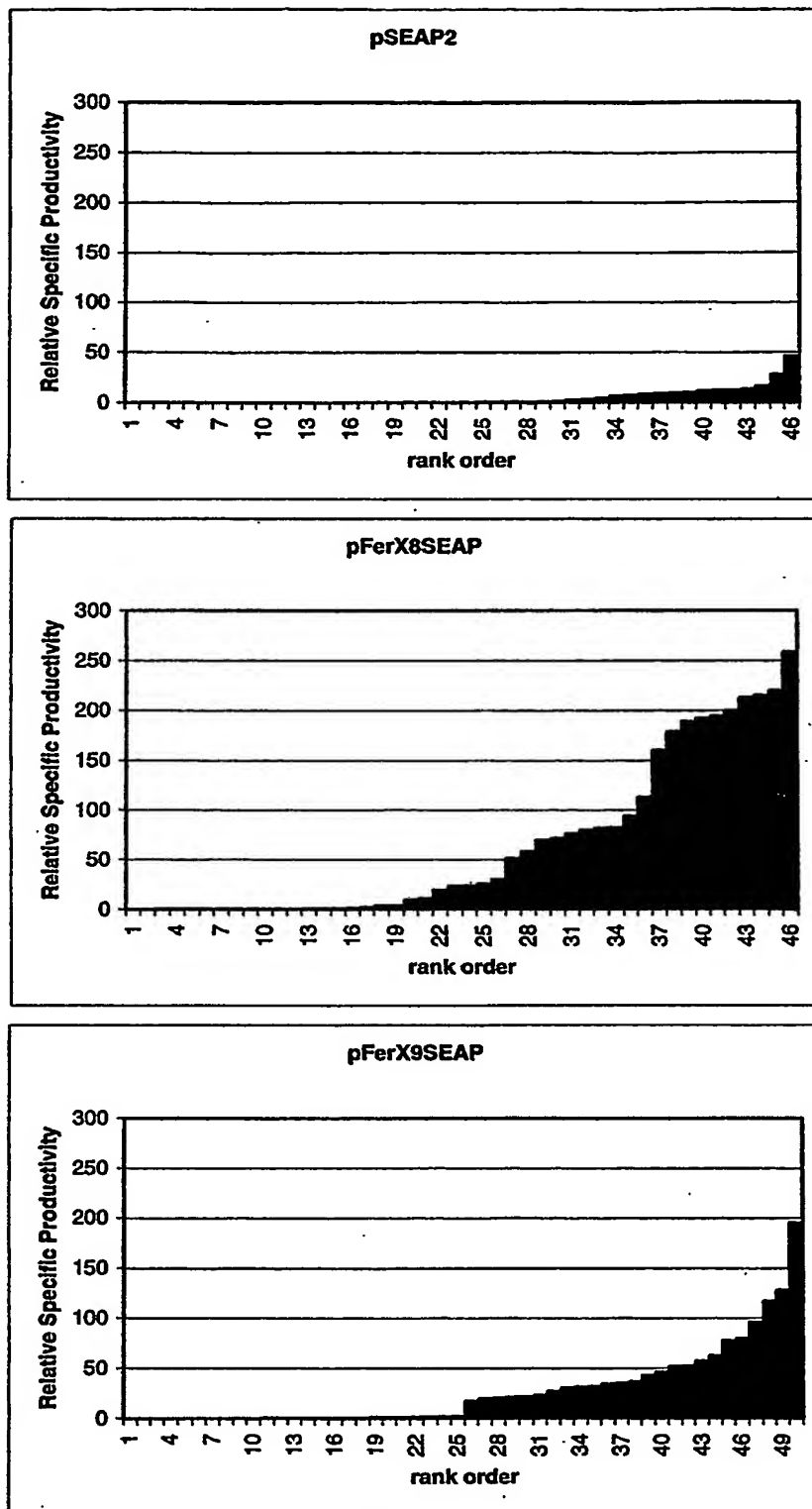
FIGURE 14

pSEAP2

pFerX8SEAP

pFerX9SEAP

FIGURE 15